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Short communication

First report of crown rot disease of guava caused by *Fusarium verticilloides* in India

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ABSTRACT

A survey was carried out in guava orchards during 2015 in Jabalpur district of Madhya Pradesh. Fruits of Thai guava were observed exhibiting initially, water-soaked lesions around the calyx region, which later enlarged into necrotic grayish-black circular patches that covered the entire fruits. The pathogen was identified as *Fusarium verticilloides*, which was confirmed by Indian Type Culture Collection, Division of Plant Pathology, ICAR-IARI, New Delhi (ID No. 9949.15). *Fusarium verticilloides* has been previously reported non-pathogenic for inducing crown rot on guava in Phillipines. This is the first report of *F. verticilloides* causing crown rot in guava in India.

Key words: Guava, crown rot, pathogenic, *Fusarium.*

Guava (*Psidium guajava* L.) is one of the dominant fruit crop of tropical and sub-tropical regions of India. Due to its hardy nature of plant, drought tolerance, has high yield potential and thus diverse uses. The fruit (berry) is an excellent source of vitamin 'C' and pectin. It is normally consumed fresh as a dessert fruit, excellent salad and puddings are prepared from the ripe fruit (Jagtiani *et al*., 4). Guava jelly is well known best jelly, besides jam, sherbet, ice cream, cheese, canned fruit, RTS, nectar, squash and powder are also prepared. Two types of wines, *viz.* guava juice and guava pulp wines are also prepared from ripe fruits (Bardiya *et al*., 2). The medicinal properties of guava fruit, leaves, and other plant parts are also well-known in the traditional medicine (Joseph and Mini, 5).

However, guava fruits ripen rapidly and are perishable due to their climacteric nature. Fruit ripening in guava is characterized by loss of green colour, softening, shrinkage, loss of brightness and rot development (Bassetto *et al*., 3; Krishna and Rao, 6). In a survey carried out during 2015 in Jabalpur district of Madhya Pradesh (India), guava fruits (variety Thai guava) were observed exhibiting initially, water-soaked lesions around the calyx region, which later enlarged into necrotic greyish-black circular patches that covered the entire fruits (Fig. 1). When infected fruits were kept in humid *in vitro* conditions, whitish-cotton like growth was observed, which developed very fast as the fruit matured and pathogen could cover almost the entire surface within 3-4 days. Under high relative humidity, the fruits near the soil level covered with dense foliage were most severely affected. The fallen fruits were also badly affected. The peel of the fruit below the whitish cottony growth becomes soft, turned light pink to dark. The objective of this study was to describe the disease symptoms and to identify the causal fungus based on morphological, cultural and pathogenic characteristics.

Fig. 1. Natural disease occurrence on guava.

Infected portion of guava fruits were cut into small fragments (1-2 cm) and surface-sterilized in 0.1% mercuric chloride for 30 sec. They were then washed three times in sterile distilled water, plated on potato dextrose agar (PDA) medium containing streptomycin sulphate and incubated at room temperature (28 ± 2°C) for seven days. Single fungal spore isolates, which were consistently recovered from infected tissues were maintained on PDA slants. Phytopathogenicity test was guided by the Koch's postulates (Agrios, 1), which state that the pure cultures of the organisms must produce the symptoms and signs of the disease when inoculated

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into the healthy plant and the suspected causal organism must be re-isolated in pure culture from the inoculated plant and must be identical to the original organism. Spore suspensions of fungal isolates were prepared individually using a PDA and standardized to 5 × 10^7 spores/ ml using haemocytometer. Drop inoculation technique (Singh, 7) was used for infecting the guava fruit. Artificial wounds were made using sterile blood lancets. Each guava fruit was pricked 30 times near the crown and 1 ml of fungal inoculum was inoculated on the artificial wounds. For the control, guava fruits were inoculated with sterile double-distilled water. Guava fruits were placed in clean culture bottles lined with moistened tissue paper to maintain the humidity inside the bottle. Then, culture bottles were covered with cheese cloth and were incubated at 28-30°C for 7 days. Fungi were re-isolated from the advancing margin of the infected tissue of the guava fruit and were inoculated in PDA plates. Cultures were incubated at 28-30°C for 5 to 7 days. The characters of the re-isolated pathogens were compared with their original isolates. This was done to prove that the isolated organism is the same organism that has been inoculated causing the specific disease. Identification was based on cultural and morphological characteristics of the fungal isolates. Symptoms similar to those observed in the field were detected two weeks post inoculation and a fungus that was morphologically similar to the inoculated culture was re-isolated from lesions of the artificially inoculated fruit (Fig. 2). Colonies on PDA were initially white and later became pink. Macroconidia were straight; spindle as well as sickle shaped and had 1-6 septa. The size of macro conidia was 15.46-44.28 µm × 4.91-9.14 µm (Fig. 3). The microconidia were hyaline, round to oval in shape and had 0-1 septa. The size of microconidia was 3.57-14.28 µm × 2.68-4.46 µm chlamydospores were also found and were round, oval, terminal and intercalary in all the isolates (Table 1). The size of chlamydospores varied from 6.85-7.73 µm × 6.67-7.90 µm. Based on cultural and morphological

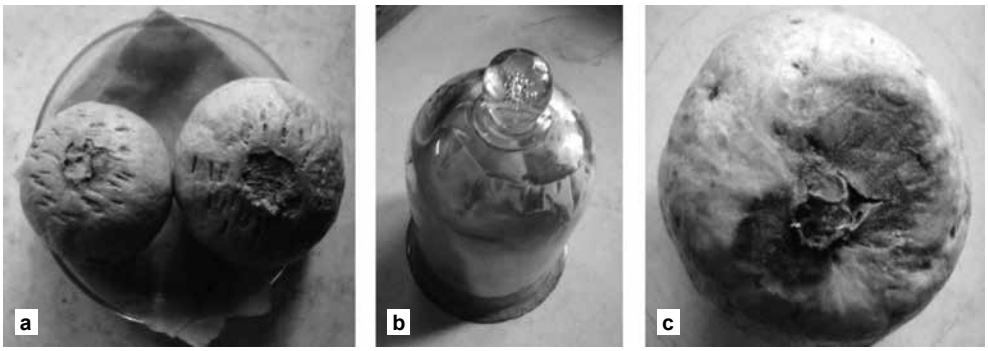

Fig. 2. Pathogenicity test on (A) Artificial wounds of guava (*in vitro*), and (B) Glass jar conditions, and (c) expression of symptoms.

Table 1. Cultural and morphological characterization *Fusarium verticilloides*.

Cultural and morphological characterization	*Fusarium verticilloides* parameter / description
Colony diameter (196 h) (mm)	78.3
Colony type	Pink cottony and fluffy growth
Color colour	Pink
Colony margin	Regular, pink with white rim
Colour on the underside of petri-plate	Brown
Macroconidia morphology	Spindle as well as sickle shaped
Mean length and width of macroconidia (µm)	15.46-44.28 × 4.91-9.14
Mean septation of macroconidia	1-6 septa
Microconidia morphology	Round to oval
Mean length & width of microconidia (µm)	3.57-14.28 × 2.68-4.46
Mean septation of microconidia	0-1 septa
Size of chlamydospores (µm)	6.85-7.73 × 6.67-7.90

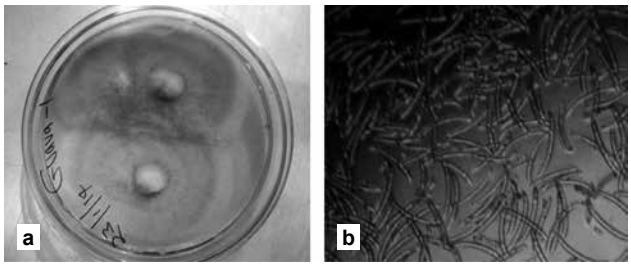

Fig. 3. Morphology of the *Fusarium verticilloides* (a) Colony of *F. verticilloides* cultured on PDA plates at 25°C for seven days (b) Macro-conidia of *F. verticilloides.*

characters, the fungus was identified as *Fusarium verticilloides,* which was confirmed by Indian Type Culture Collection, ICAR-IARI, New Delhi (I.D. No. 9949.15). *Fusarium verticilloides* has been previously reported non-pathogenic to crown rot on guava in Phillipines (Valentino *et al*., 8). This is the first report of *F. verticilloides* causing crown rot on guava in India.

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